# Product Information

## 6-CDCFDA, SE [Carboxy-2',7'-dichlorofluorescein diacetate, succinimidyl ester]

Catalog Number: C4069

Product Size: 5 mg

Application Scope: Cell viability probe

## Parameters

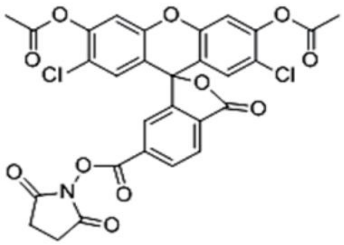
Appearance: White solid soluble in DMSO or DMF

Ex/Em: 492/517 nm

Molecular Formula: $C_{29}H_{17}Cl_2NO_{11}$

Molecular Weight: 626

Molecular Structure:



## Storage

Store at -20℃ and protect from light. When stored as directed, product is stable for at least 12 months.

## Description

6-CDCFDA, SE is a live cell fluorescent tracer probe with membrane permeability and thus can be loaded into cells via incubation. It can be used for in vivo and in vitro experiments on cell proliferation.

6-CDCFDA, SE is a derivative of Fluorescein diacetate (FDA), which has cell membrane permeability and does not have fluorescence itself. Once inside cells, the non-fluorescent CFDA is hydrolyzed by intracellular esterases to carboxyfluorescein succinimidyl ester (CFSE). These fluorescent products can only accumulate in cells with integral cell-membrane and can emit strong green fluorescence. CFSE can spontaneously and irreversibly bind to intracellular amino groups to couple to cellular proteins. The unconjugated 6-CDCFDA, SE is returned to the cell by passive diffusion and is removed by subsequent washing steps. The fluorescence of SE-labeled cells can be stably labeled for several months, so it is very suitable for cell community analysis.

The fluorescence of 6-CDCFDA, SE-labeled cells is very uniform, which is better than other cell-tracking fluorescent probes such as PKH26. In the process of cell division and proliferation, CFSE-labeled fluorescence can be evenly distributed to the two progeny cells, the fluorescence intensity becomes half that of the parent cell, and non-dividing and dividing cells can be detected according to the difference in fluorescence intensity. Cell division once, fluorescence intensity becomes 1/2; Cell division twice, fluorescence intensity becomes 1/4; Cell division three times, fluorescence intensity becomes 1/8. 6-CDCFDA, SE can detect splits up to eight times. 6-CDCFDA, SE-labeled cells can be used for in vitro and in vivo proliferation studies, and have the function of not staining neighboring cells. 6-CDCFDA, SE can be used to detect the proliferation of lymphocytes, fibroblasts, natural killer cells, hematopoietic progenitor cells and other cells.

6-CDCFDA, SE can not only detect cell proliferation by flow cytometry, but also quantify the number of living cells with a fluorescence microplate, or use a fluorescence microscope for uniformly stained cell trace observation.



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## Protocol

The following are the staining steps for live cells, which can be adjusted as appropriate.

Note: 6-CDCFDA, SE reacts with amine groups. Do not use amine-containing buffers in experiments.

1. A 10 mM stock solution can be configured using anhydrous DMSO and diluted to a working concentration of 0.5-25 μM with PBS.

Note: It is recommended to use 6-CDCFDA, SE working solution immediately after configuration.

Note: The specific incubation concentration and incubation time are different depending on the cells. It is recommended to set a set of gradients to explore the best experimental conditions. If the cells divide faster or stay longer after staining, it is recommended to increase the working concentration to 5-10 μM.

2. Collected cells by centrifugation, and resuspended cells in 6-CDCFDA, SE working solution pre-warmed at 37 ° C.

3. Incubate cells for 15~30 minutes at 37°C.

4. Discard 6-CDCFDA, SE working solution and wash the cells twice with PBS.

5. Analyze fluorescence by fluorescence microscopy or flow cytometry.

The following are optional steps (if antibody labeling is required, fixation and permeabilization can be performed):

6. Fixed. It can be fixed for 15 min at room temperature using 3.7% paraformaldehyde.

7. Permeability. Permeabilize in ice acetone for 10 min.

8. After fixation and permeabilization, cells need to be washed with PBS.

## Notes

1. 6-CDCFDA, SE reacts with amine groups. Do not use amine-containing buffers in experiments.

2. There are quenching problems with fluorescent dyes. Please avoid light to slow down the fluorescence quenching.

3. For your safety and health, please wear lab coats and disposable gloves.



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